

WHAT IS CLAIMED IS:

1. A portable DNA sequence comprising a series of nucleotides capable of directing intracellular production of metalloproteinase inhibitors.

2. The portable DNA sequence of claim 1 wherein said sequence is capable of directing intracellular production of collagenase inhibitors.

3. The portable DNA sequence of claim 1 wherein said nucleotide sequence is:

10	20	30	40	50	60
GTTGTTGCTG	TGGCTGATAG	CCCCAGCAGG	GCCTGCACCT	GTGTCCCACC	CCACCCACAC
70	80	90	100	110	120
ACGGCCTTCT	GCAATTCCGA	CCTCGTCATC	AGGGCCAAGT	TCGTGGGGAC	ACCAGAAGTC
130	140	150	160	170	180
AACCAGACCA	CCTTATACCA	GCGTTATGAG	ATCAAGATGA	CCAAGATGTA	TAAAGGGTTG
190	200	210	220	230	240
CAAGCCTTAG	GGGATGCCGC	TGACATCCGG	TTCGTCTACA	CCCCCGCCAT	GGAGAGTGTG
250	260	270	280	290	300
TGCGGATACT	TCCACAGGTC	CCACAACCGC	AGCGAGGAGT	TTCTCATTCG	TGGAAAACCT
310	320	330	340	350	360
CAGGATGGAC	TCTTGACAT	CACTACCTGC	AGTTTCGTGG	CTCCCTGGAA	CAGCCTGAGT
370	380	390	400	410	420
TTAGCTCAGC	GCCGGGGCTT	CACCAAGACC	TACACTGTTG	GCTGTGAGGA	ATGCACAGT
430	440	450	460	470	480
TTTCCCTGTT	TATCCATCCC	CTGCAAACTG	CAGAGTGGCA	CTCATTGCTT	GTGGACGGA
490	500	510	520	530	540
CAGCTCCTCC	AAGGCTCTGA	AAAGGGCTTC	CAGTCCCGTC	ACCTTGCCTG	CCTGCCTCG
550	560	570	580	590	600
GAGCCAGGGC	TGTGCACCTG	GCAGTCCCTG	CGGTCCCAGA	TAGCCTGAAT	CCTGCCCCGG

610	620	630	640	650	660
GTGGAAGCTG	AAGCCTGCAC	AGTGTCCACC	CTGTTCCCAC	TCCCATCTTT	CTTCCGGACA
670	680	690	700		
ATGAAATAAA	GAGTTACCAC	CCAGCAAAAA	AAAAAAGGAA	TTC	

4. The portable DNA sequence of claim 2 wherein said sequence is capable of directing intracellular production of a collagenase inhibitor biologically equivalent to that isolable from human skin fibroblasts.

5. A recombinant-DNA cloning vector comprising a nucleotide sequence capable of directing intracellular production of metalloproteinase inhibitors.

6. The vector of claim 5 wherein said vector comprises a nucleotide sequence containing at least the following nucleotides:

10	20	30	40	50	60
GTTGTTGCTG	TGGCTGATAG	CCCCAGCAGG	GCCTGCACCT	GTGTCCCACC	CCACCCACAG
70	80	90	100	110	120
ACGGCCTTCT	GCAATTCCGA	CCTCGTCATC	AGGGCCAAGT	TCGTGGGGAC	ACCAGAAGTC
130	140	150	160	170	180
AACCAGACCA	CCTTATACCA	GCGTTATGAG	ATCAAGATGA	CCAAGATGTA	TAAAGGGTTC
190	200	210	220	230	240
CAAGCCTTAG	GGGATGCCGC	TGACATCCGG	TTCGTCTACA	CCCCCGCCAT	GGAGAGTGTC
250	260	270	280	290	300
TGCGGATACT	TCCACAGGTC	CCACAACCGC	AGCGAGGAGT	TTCTCATTGC	TGGAAAAGTG
310	320	330	340	350	360
CAGGATGGAC	TCTTGACAT	CACTACCTGC	AGTTTCGTGG	CTCCCTGGAA	CAGCCTGAGC
370	380	390	400	410	420
TTAGCTCAGC	GCCGGGGCTT	CACCAAGACC	TACACTGTTG	GCTGTGAGGA	ATGCACAGTG
430	440	450	460	470	480
TTTCCCTGTT	TATCCATCCC	CTGCAAAGTG	CAGAGTGGCA	CTCATTGCTT	GTGGACGGAC

490	500	510	520	530	540
CAGCTCCTCC	AAGGCTCTGA	AAAGGGCTTC	CAGTCCCGTC	ACCTTGCCTG	CCTGCCTCGC
550	560	570	580	590	600
GAGCCAGGGC	TGTGCACCTG	GCAGTCCCTG	CGGTCCCAGA	TAGCCTGAAT	CCTGCCCCGA
610	620	630	640	650	660
GTGGAAGCTG	AAGCCTGCAC	AGTGTCCACC	CTGTTCCCAC	TCCCATCTTT	CTTCCGGACA
670	680	690	700		
ATGAAATAAA	GAGTTACCAC	CCAGCAAAAA	AAAAAAGGAA	TTC	

7. The vector pUC9-F5/237P10.

8. A recombinant-DNA method for microbial production of a metalloproteinase inhibitor comprising:

- (a) preparation of a portable DNA sequence capable of directing a host microorganism to produce a protein having metalloproteinase inhibitor activity;
- (b) cloning the portable DNA sequence into a vector capable of being transferred into and replicating in a host microorganism, such vector containing operational elements for the portable DNA sequence;
- (c) transferring the vector containing the portable DNA sequence and operational elements into a host microorganism capable of expressing the metalloproteinase inhibitor protein;
- (d) culturing the host microorganism under conditions appropriate for amplification of the vector and expression of the inhibitor; and
- (e) in either order:
 - (i) harvesting the inhibitor; and
 - (ii) causing the inhibitor to assume an active, tertiary structure whereby it possesses metalloproteinase inhibitor activity.

9. The method of claim 8 wherein said metalloproteinase inhibitor is collagenase inhibitor.

10. The method of claim 8 wherein said portable DNA sequence is:

10	20	30	40	50	60
GTTGTTGCTG	TGGCTGATAG	CCCCAGCAGG	GCCTGCACCT	GTGTCCCACC	CCACCCACAG
70	80	90	100	110	120
ACGGCCTTCT	GCAATTCCGA	CCTCGTCATC	AGGGCCAAGT	TCGTGGGGAC	ACCAGAAGTC
130	140	150	160	170	180
AACCAGACCA	CCTTATACCA	GCGTTATGAG	ATCAAGATGA	CCAAGATGTA	TAAAGGGTTC
190	200	210	220	230	240
CAAGCCTTAG	GGGATGCCGC	TGACATCCGG	TTCGTCTACA	CCCCCGCCAT	GGAGAGTGTC
250	260	270	280	290	300
TGCGGATACT	TCCACAGGTC	CCACAACCGC	AGCGAGGAGT	TTCTCATTGC	TGGAAAACTG
310	320	330	340	350	360
CAGGATGGAC	TCTTGACAT	CACTACCTGC	AGTTTCGTGG	CTCCCTGGAA	CAGCCTGAGC
370	380	390	400	410	420
TTAGCTCAGC	GCCGGGGCTT	CACCAAGACC	TACACTGTTG	GCTGTGAGGA	ATGCACAGTG
430	440	450	460	470	480
TTTCCCTGTT	TATCCATCCC	CTGCAAACCTG	CAGAGTGGCA	CTCATTGCTT	GTGGACGGAC
490	500	510	520	530	540
CAGCTCCTCC	AAGGCTCTGA	AAAGGGCTTC	CAGTCCCGTC	ACCTTGCCTG	CCTGCCTCGG
550	560	570	580	590	600
GAGCCAGGGC	TGTGCACCTG	GCAGTCCCTG	CGGTCCCAGA	TAGCCTGAAT	CCTGCCCCGA
610	620	630	640	650	660
GTGGAAGCTG	AAGCCTGCAC	AGTGTCCACC	CTGTTCCCAC	TCCCATCTTT	CTTCCGGACA
670	680	690	700		
ATGAAATAAA	GAGTTACCAC	CCAGCAAAAA	AAAAAAGGAA	TTC	

11. The method of claim 8 wherein said vector containing said portable DNA sequence is pUC9-F5/237P10.

12. The method of claim 8 wherein said host microorganism is a bacterium.

13. The method of claim 12 wherein said bacterium is a member of the genus Bacillus.

14. The method of claim 13 wherein said bacterium is Bacillus subtilis.

15. The method of claim 12 wherein said bacterium is Escherichia coli.

16. The method of claim 12 wherein said bacterium is a member of the genus Pseudomonas.

17. The method of claim 16 wherein said bacterium is Pseudomonas aeruginosa.

18. The method of claim 8 wherein said host microorganism is a yeast.

19. The method of claim 8 wherein said yeast is Saccharomyces cerevisiae.

20. The method of claim 8 wherein said inhibitor is harvested prior to being caused to assume said active, tertiary structure.

21. The method of claim 8 wherein said inhibitor is caused to assume said active, tertiary structure prior to being harvested.

22. A metalloproteinase inhibitor which is biologically equivalent to the collagenase inhibitor isolable from human skin fibroblasts produced by the method of claim 8.

23. The microorganism C600/pUC9-F5/237P10 having ATCC
Accession No. 53003.

24. The portable DNA sequence of claim 1 wherein said
nucleotide sequence is:

10	20	30	40	50	60
GGCCATCGCC	GCAGATCCAG	CGCCCAGAGA	GACACCAGAG	AACCCACCAT	GGCCCCCTT
70	80	90	100	110	120
GACCCCTGGC	TTCTGCATCC	TGTTGTTGCT	GTGGCTGATA	GCCCCAGCAG	GGCCTGCAC
130	140	150	160	170	180
TGTGTCCCAC	CCCACCCACA	GACGGCCTTC	TGCAATTCCG	ACCTCGTCAT	CAGGGCCAA
190	200	210	220	230	240
TTCGTGGGGA	CACCAGAAGT	CAACCAGACC	ACCTTATACC	AGCGTTATGA	GATCAAGAT
250	260	270	280	290	300
ACCAAGATGT	ATAAAGGGTT	CCAAGCCTTA	GGGGATGCCG	CTGACATCCG	GTTTCGTCTA
310	320	330	340	350	360
ACCCCCGCCA	TGGAGAGTGT	CTGCGGATAC	TTCCACAGGT	CCCACAACCG	CAGCGAGGAC
370	380	390	400	410	420
TTTCTCATTG	CTGGAAACT	GCAGGATGGA	CTCTTGACAC	TCACTACCTG	CAGTTTCGTC
430	440	450	460	470	480
GCTCCCTGGA	ACAGCCTGAG	CTTAGCTCAG	CGCCGGGGCT	TCACCAAGAC	CTACACTGTT
490	500	510	520	530	540
GGCTGTGAGG	AATGCACAGT	GTTTCCCTGT	TTATCCATCC	CCTGCAAAC	GCAGAGTGGC
550	560	570	580	590	600
ACTCATTGCT	TGTGGACGGA	CCAGCTCCTC	CAAGGCTCTG	AAAAGGGCTT	CCAGTCCCCT
610	620	630	640	650	660
CACCTTGCCT	GCCTGCCTCG	GGAGCCAGGG	CTGTGCACCT	GGCAGTCCCT	GCGGTCCCAG
670	680	690	700	710	720
ATAGCCTGAA	TCCTGCCCCG	AGTGGAAGCT	GAAGCCTGCA	CAGTGTCCAC	CCTGTTCCCA
730	740	750	760	770	780
CTCCCATCTT	TCTTCCGGAC	AATGAAATAA	AGAGTTACCA	CCCAGCAAAA	AAAAAAGGA